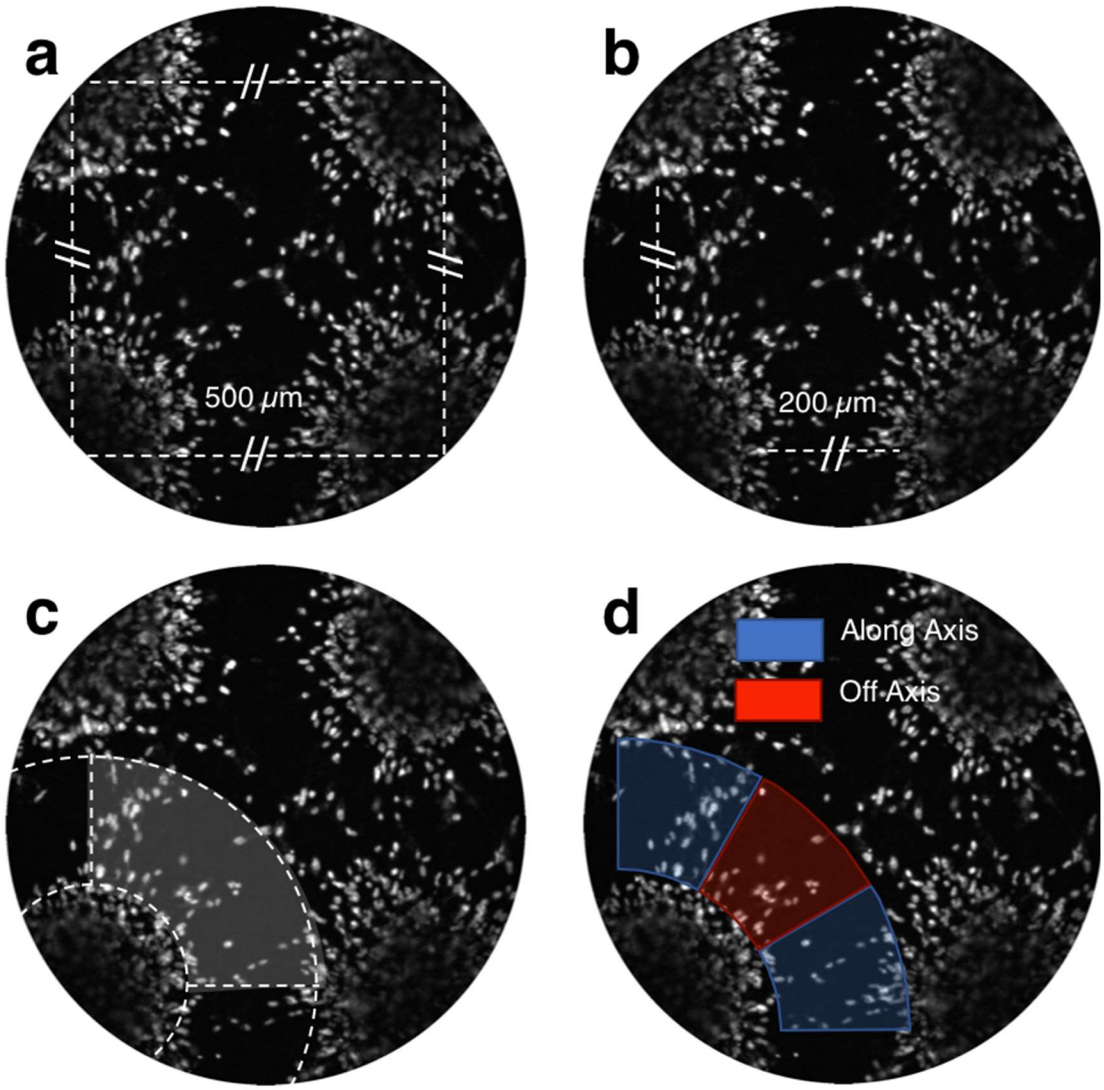
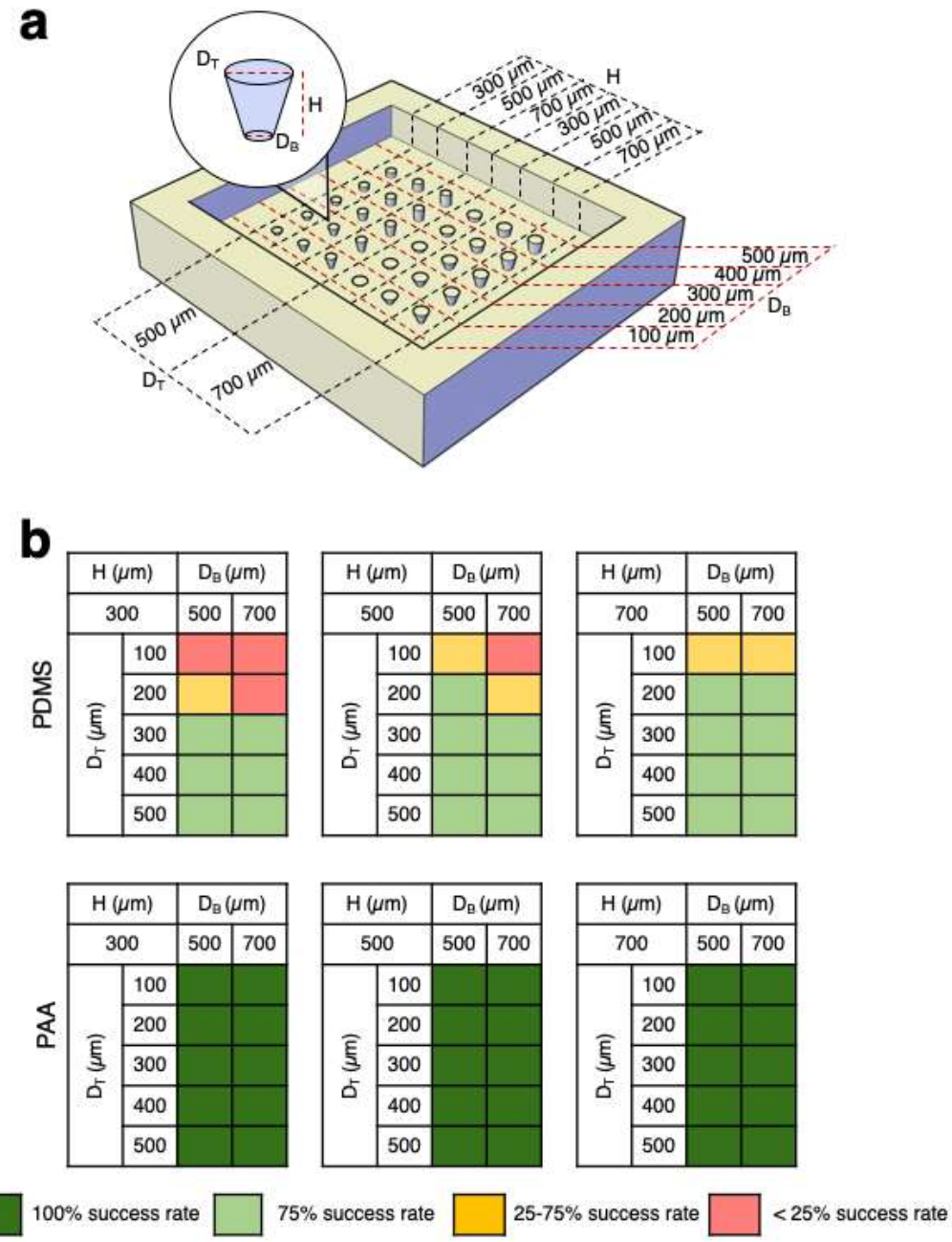
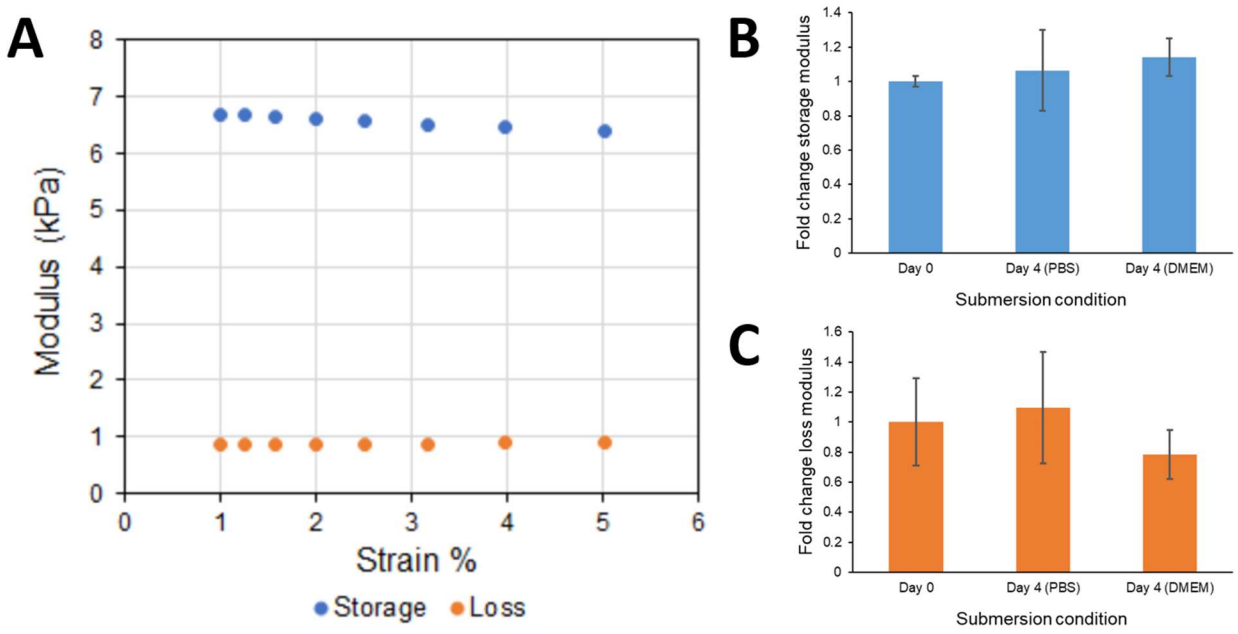
Supplemental Figures



Supplemental Figure 1. Defining regions of interest for cell invasion and cell orientation quantification. **(a)** Micropockets were spaced 500 μm apart, from center-to- center. **(b)** The spacing from edge to edge was 200 μm. **(c)** Quadrants were drawn around each of the analyzed micropockets, and **(d)** Three equal-area segments, defined as regions that were “along axis” or “off axis” were established.



Supplemental Figure 2. Determining the replicability of PDMS and PAAm. (a) To determine whether to use PDMS or PAAm for the spherical microwell platform, each material's ability to replicate reverse-tapered structures was evaluated. A mold of differently sized reverse-tapered frustums was designed in AutoCAD and 3D printed with the AutoDesk Ember DLP 3D printer. The reverse-tapered frustums were then replica molded either with softened PDMS (75 wt.% hard PDMS, 25 wt.% soft PDMS) and PAAm (25.6 kPa stiffness). **(b)** The most suitable material for experiments was based on which material had the most reliable and accurate replicability. The resulting inverse molds as well as the original molds were analyzed for defects. Replicability of the material was qualitatively based on whether there was damage to the mold (i.e. structures broken off) or damage to the replica. With this definition, replicability phase diagrams were constructed for both PDMS and PAAm.



Supplemental Figure 3. Mechanical characterization of selected polyacrylamide formulation by shear rheometry. (A) A parallel plate shear rheometer (Anton-Paar, MCR302) was used to characterized pre-gelled and swollen PAAm gels. Gels were cast between two 12-mm circular coverslips to an approximate gel height of 1 mm, allowed to swell in PBS overnight, and affixed between the parallel plates of the instrument using double sided tape, following standard and previously described protocols[47]. Shear and loss modulus values were obtained over a strain sweep from 1 to 5% strain at 10 Hz. (B, C) To confirm that PAAm hydrogels do not significantly change mechanical properties over time in culture, samples were mechanically characterized at Day 0, and after 4 days at cell culture incubator conditions (37 °C, 5% CO_2) in either PBS or DMEM. No significant differences were found in (B) storage modulus, or (C) loss modulus over this time period, consistent with previous studies demonstrating the stability of polyacrylamide gels to cell culture conditions[47] ($n = 3$, one-way ANOVA, $p > 0.47$).